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Research Article

Correlations of plasma renin activity and aldosterone concentration with ambulatory blood pressure responses to nebivolol and valsartan, alone and in combination, in hypertension

Thomas D. Giles, MD[a,*], George Bakris, MD[b], Suzanne Oparil, MD[c], Michael A. Weber, MD[d], Huiling Li, MD[e], Madhuja Mallick, PhD[e], David B. Bharucha, MD, PhD[f], ChunLin Chen, PhD[f], and William G. Ferguson, PhD[f] On behalf of the NAC-MD-01 Substudy Investigators

[a]*Department of Medicine, Tulane University, New Orleans, LA, USA;*
[b]*Department of Medicine, The University of Chicago, Chicago, IL, USA;*
[c]*Division of Cardiovascular Disease, University of Alabama School of Medicine, Birmingham, AL, USA;*
[d]*Division of Cardiovascular Medicine, Downstate College of Medicine, State University of New York, Brooklyn, NY, USA;*
[e]*Department of Biostatistics, Forest Research Institute, Inc, an affiliate of Actavis, Inc (now: Allergan), Harborside Financial Center, Jersey City, NJ, USA; and*
[f]*Department of Clinical Development, Forest Research Institute, Inc, an affiliate of Actavis, Inc (now: Allergan), Harborside Financial Center, Jersey City, NJ, USA*

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Abstract

After demonstration of the antihypertensive efficacy of the combination of the beta-blocker nebivolol and the angiotensin receptor blocker valsartan in an 8-week, randomized, placebo-controlled trial (N = 4161), we now report the effects of this treatment on the renin-angiotensin-aldosterone system in a substudy (n = 805). Plasma renin activity increased with valsartan (54%–73%) and decreased with nebivolol (51%–65%) and the combination treatment (17%–39%). Plasma aldosterone decreased with individual treatments (valsartan, 11%–22%; nebivolol, 20%–26%), with the largest reduction (35%) observed with maximum combination dose (20 mg nebivolol/320 mg valsartan). Baseline ln(plasma renin activity) correlated with the 8-week reductions in 24-hour systolic and diastolic BP following treatments with the combination (all doses combined, $P = .003$ and $P < .001$) and nebivolol (both, $P < .001$), but not with valsartan. Baseline ln(aldosterone) correlated with 24-hour systolic and diastolic BP reductions following combination treatment only ($P < .001$ and $P = .005$). The implications of the renin-angiotensin-aldosterone system effects of this beta blocker-angiotensin receptor blocker combination should be explored further. J Am Soc Hypertens 2015;9(11):845–854. © 2015 The Authors. Published by Elsevier Inc. on behalf of American Society of Hypertension. This is an open access article under the CC BY-NC-ND license (http://creativecommons.org/licenses/by-nc-nd/4.0/).

Keywords: ABPM; angiotensin II receptor blocker; biomarkers; Vasodilatory beta-blocker.

HL and CLC are no longer affiliated with Forest Research Institute.

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Conflict of interest: In the past two years, T.D. Giles has received personal fees from Forest Laboratories (now: Actavis). G. Bakris is an advisor or consultant for AbbVie, Bayer, BMS, CVRx, Elcelyx, Eli Lilly/Boehringer-Ingelheim, GSK, Janssen, Medtronic, Novartis, Takeda, Tegion, and ZS Pharma. S. Oparil has received research support from Amarin Pharma, AstraZeneca, Bayer, Daiichi-Sankyo, LipoScience, Medtronic, Ardian, and Merck, and served as a consultant or advisory board member for AstraZeneca, Bayer, Daiichi-Sankyo, Forest, Medtronic, Ardian, Novartis, and Takeda. M.A. Weber has received speaker, consultant, or research fees from Arbor, AstraZeneca, Boston Scientific, Daiichi-Sankyo, Forest, Medtronic, and Takeda. H. Li, M. Malick, D.B. Bharucha, C.L. Chen, and W.G. Ferguson are employees, present (M. Malick, D.B. Bharucha, W.G. Ferguson) or former (H. Li, C.L. Chen), of Forest Research Institute, an affiliate of Actavis Inc. (now: Allergan), the US marketer of nebivolol.

*Corresponding author: Thomas D. Giles, MD, Tulane University, 109 Holly Drive, Metairie, LA 70005, USA. Tel: +1-504-834-8668; Fax: +1-504-836-0822.

E-mail: tgiles4@cox.net

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Introduction

Plasma renin activity (PRA) was one of the earlier biomarkers of hypertension and drew attention to the role of the renin-angiotensin-aldosterone system (RAAS) in the pathophysiology of hypertension.[1] Renin, produced in the juxtaglomerular cells of the kidney, is the rate-limiting enzyme in the renin-angiotensin system and is responsible for converting angiotensinogen to angiotensin I.[1] Angiotensin I is subsequently processed by the angiotensin I-converting enzyme to angiotensin II, the main effector peptide of the system.[1]

The emergence of antihypertensive drugs that perturb the RAAS provided an opportunity to further explore the system's role in blood pressure (BP) regulation. It was reasoned that patients with higher baseline PRAs would have a greater reduction in BP to renin-angiotensin-aldosterone system-blocking drugs than patients with lower PRA. In addition, it was proposed that PRA could be useful in characterizing patients with hypertension who had vasoconstriction as the principal mechanism responsible for BP elevation. This strategy has been tested many times, but with inconsistent results.[2,3]

A recently completed large randomized trial (N = 4161) in individuals with hypertension demonstrated that an antihypertensive single-pill combination (SPC) consisting of nebivolol, a β_1-selective vasodilatory β-blocker,[4] and valsartan,[5] an angiotensin II receptor blocker (ARB), is more efficacious in reducing BP than the individual drugs.[6] In a substudy of that trial (n = 805), we measured PRA and aldosterone concentrations, providing us with an opportunity to examine whether these measurements could help explain the effects of treatment on BP measured by ambulatory monitoring. In the following we report the results of that substudy.

Methods

Study Design

The details of study design were reported previously.[6] Briefly, the study was a phase 3, multicenter, randomized, double-blind, placebo-controlled, parallel-group, dose-titration trial (NAC-MD-01; NCT01508026) in which participants, after a 1-week screening period, entered a 6-week single-blind placebo run-in phase, followed by an 8-week double-blind treatment period. The participants were randomized (2:2:2:2:2:2:2:1) to 4 weeks of double-blind treatment with a nebivolol-valsartan SPC (5/80, 5/160, or 10/160 mg/day), nebivolol monotherapy (5 or 20 mg/day), valsartan monotherapy (80 or 160 mg/day), or placebo. Dosages were doubled at the beginning of week 5 to SPC 10/160, 10/320, or 20/320 mg/day, nebivolol 10 or 40 mg/day, or valsartan 160 or 320 mg/day. After 8 weeks of double-blind treatment, the study drug dose was tapered over a 1-week double-blind down-titration phase.

Participants

Eligible participants were men or women ≥18 years of age, diagnosed with Stage 1 or 2 hypertension (JNC7 criteria[7]), and with diastolic BP (DBP) measurements of ≥95 mm Hg and <110 mm Hg (untreated) or ≥90 mm Hg and <110 mmHg (treated) at screening, and a seated pulse rate of ≥55 bpm. Participants were also required to have normal or clinically nonsignificant results of physical examination, laboratory tests, and electrocardiogram at screening. Key exclusion criteria were secondary hypertension, severe hypertension (SBP ≥180 mm Hg or DBP ≥110 mm Hg), current treatment with ≥4 antihypertensive medications (including components of the SPC), contraindication to discontinuing antihypertensive treatment, upper arm circumference >42 cm, the presence of symptomatic coronary artery disease, reactive airways disease, chronic obstructive pulmonary disease, second- or third-degree heart block or sick sinus syndrome, type 1 diabetes, poorly controlled type 2 diabetes (HbA_{1C} ≥ 8%), uncontrolled thyroid disease within 3 months of screening, and pregnancy or breastfeeding. In addition, shift workers were excluded from the substudy.

Biomarkers-Ambulatory Blood Pressure Monitoring Substudy

Predose blood samples for the assessment of PRA and aldosterone were collected at Baseline and at weeks 4 and 8. All samples were immediately frozen in a mixture of ethanol and ice and shipped on dry ice to the central laboratory (Keystone Bioanalytical Inc., North Wales, PA, USA). PRA was determined by means of an enzyme-linked immunosorbent assay and aldosterone levels were determined using high performance liquid chromatography coupled with tandem mass spectrometry. Ambulatory blood pressure monitoring (ABPM) measurements were recorded at baseline and at the end of week 8; the devices were applied between 8 AM and 10 AM on the nondominant arm and the data were processed by BioClinica, Inc. (Princeton, NJ, USA).

Data Analysis

Changes in 24-hour DBP and SBP in all groups were prespecified as additional efficacy parameters. Analyses were performed based on the intention-to-treat (ITT) population in the ABPM substudy, using the last observation carried forward approach to impute missing data. Treatment groups were compared using an analysis of covariance model with treatment group and diabetes status as factors and baseline value as a covariate; nominal

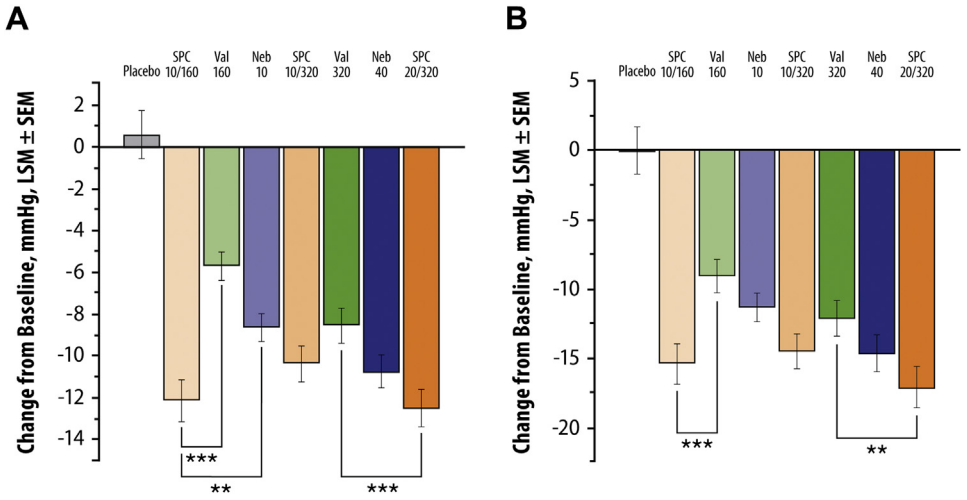

Figure 1. Baseline-to-endpoint changes in 24-hour ABPM parameters (ITT, LOCF, ANCOVA) (A) DBP; (B) SBP. $^{**}P < .01$; $^{***}P < .001$. ABPM, ambulatory blood pressure monitoring; ANCOVA, analysis of covariance; DBP, diastolic blood pressure; ITT, intent-to-treat population; LOCF, last observation carried forward; LS, least squares; LSM, least squares mean; Neb, nebivolol; Pbo, placebo; SBP, systolic blood pressure; SEM, standard error of the mean; Val, valsartan.

P-values are provided. Post hoc BP analyses with pooled active treatment groups by biomarker level quartiles (Figure 4) were performed using the same model.

Protocol-based biomarker analyses were performed by means of descriptive statistics, using the ITT population in the substudy and the last observation carried forward approach for imputation of missing data. Post hoc comparisons were performed using a general linear model with treatment group and diabetes status as class variables and log-transformed baseline values as a covariate; nominal *P*-values are provided. Post hoc correlations between 24-hour, daytime, and nighttime DBP and SBP changes in pooled active treatment groups and natural logarithm values of PRA and aldosterone concentration at baseline (Table 2) were assessed by means of Pearson's *R*.

Results

Patient Characteristics

Of 4161 randomized participants, 805 (19.3%) participated in the substudy. Baseline characteristics of the ITT population in the ABPM substudy (N = 797) are summarized in Table 1.

ABPM Analyses

From baseline to week 8, SPC 20/320 was significantly more effective than Val 320 in reducing both 24-hour DBP (least squares mean difference [LSMD] [95% CI]: −4.6 [−6.6, −2.5] mm Hg, $P < .001$) and 24-hour SBP (−5.1 [−8.4, −1.8] mm Hg, $P = .003$), but not more than Neb 40 (DBP: −1.6 [−3.7, 0.6] mm Hg, $P = .149$; SBP: −2.1 [−5.4, 1.3] mm Hg, $P = .231$; Figure 1).

Additional analyses suggest that SPC 10/160 was significantly more effective than both of its component monotherapies in reducing 24-hour DBP and more effective than Val 160 in reducing 24-hour SBP (Figure 1).

Biomarker Analyses

The geometric mean PRA values at baseline and endpoint are presented in Figure 2A. After 8 weeks of double-blind treatment, PRA levels in the groups treated with Val 160 and Val 320 increased by 53.8% and 72.8%, respectively, and decreased in the groups treated with nebivolol (by 51.3% [10 mg/day] and 65.4% [40 mg/day]) and the SPCs (from 3.2% to 39.0%; Figure 2B). A post hoc analysis revealed significant differences in PRA geometric mean ratios between SPCs 20/320 and 10/320 versus their monotherapy components, plus SPC 10/160 versus Val 160 (Figure 2B).

The geometric mean aldosterone concentrations at baseline and endpoint are summarized in Figure 3A. After 8 weeks of double-blind treatment, aldosterone level in the placebo group increased by 17.1%, compared with the baseline; in all other groups aldosterone levels decreased, from 11.1% to 35.1%. The greatest decrease was in the SPC 20/320 group, which exceeded the reductions observed in the Neb 40 group and the Val 320 group by 32.8% and 59.0%, respectively (Figure 3B). A post hoc analysis revealed that the difference in aldosterone change from baseline between SPC 20/320 and Val 320 was statistically significant (Figure 3B).

Post hoc analysis of pooled active treatment groups revealed that natural logarithm (ln) baseline levels of PRA were significantly correlated with 24-hour and daytime BP changes at week 8 in participants treated

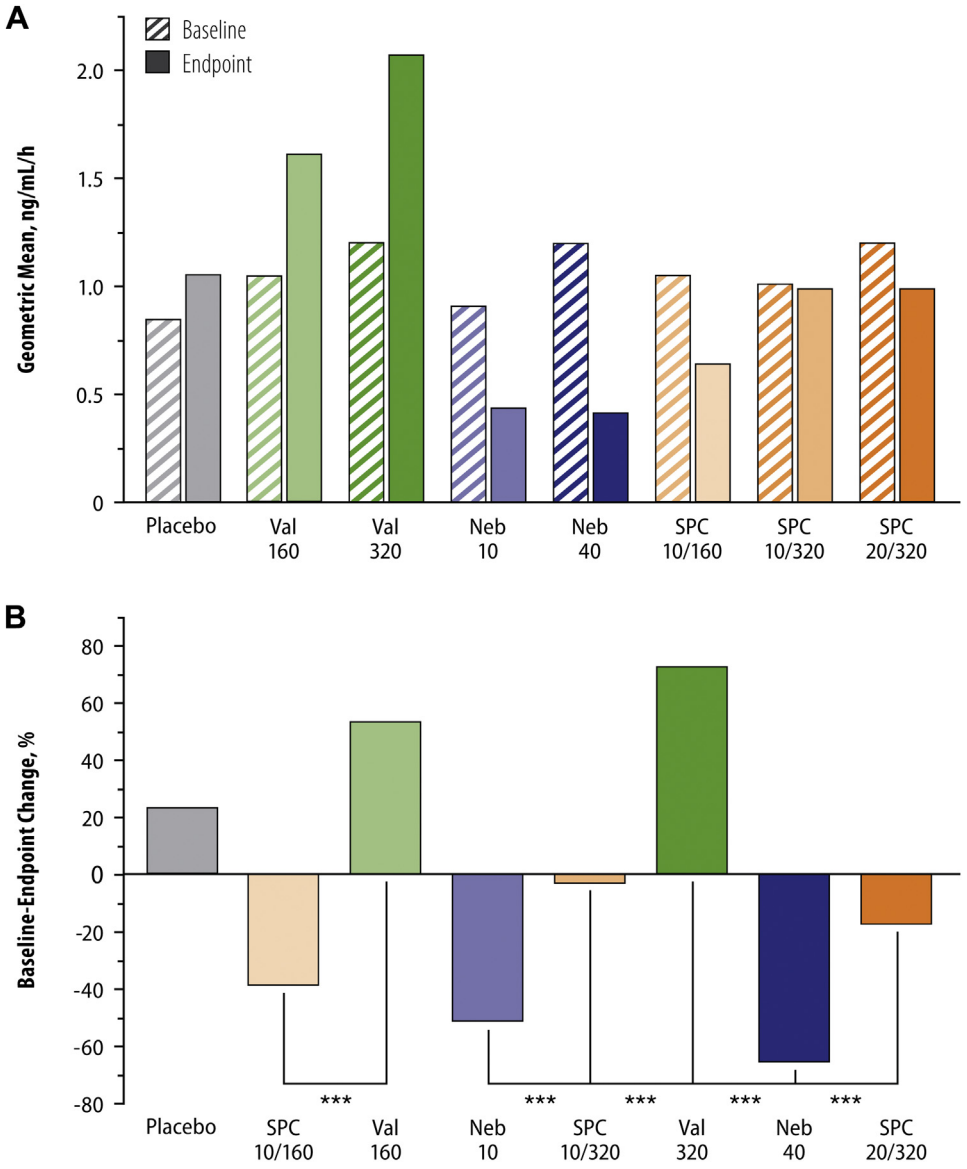

Figure 2. PRA levels and changes from baseline (ITT, LOCF) (A) Baseline and Endpoint Concentrations; (B) Baseline-to-Endpoint Change. ***$P < .001$. ITT, intent-to-treat population; LOCF, last observation carried forward; Neb, nebivolol; PRA, plasma renin activity; SPC, single-pill combination of nebivolol and valsartan; Val, valsartan.

with nebivolol and the SPCs, but not valsartan (Table 2). In addition, 24-hour, daytime, and nighttime BP changes (except nighttime DBP) were significantly correlated with natural logarithm baseline levels of aldosterone in participants treated with the SPCs, but not with the monotherapies (Table 2).

Twenty-four-hour DBP changes by baseline quartiles of PRA and aldosterone levels are shown in Figure 4.

Discussion

As expected, administration of valsartan monotherapy resulted in a dose-dependent increase in PRA, which is a well-documented effect of ARB treatment.[8,9] In contrast, nebivolol monotherapy decreased PRA in a dose-dependent manner, which is consistent with previously published data on nebivolol and other β-blockers.[10,11] As hypothesized, the nebivolol-valsartan combination was associated with a reduction in PRA, which is likely a result of nebivolol-induced attenuation of the valsartan-induced increase, and consistent with results of a single-dose study in healthy adult men given monotherapy or combination therapy consisting of valsartan and the β_1-selective antagonist atenolol.[12] To the best of our knowledge, our study is the first to assess the combined effect of a β-blocker and an ARB on PRA and aldosterone, and the first to assess the antihypertensive effect of that combination in the context of baseline biomarker levels.

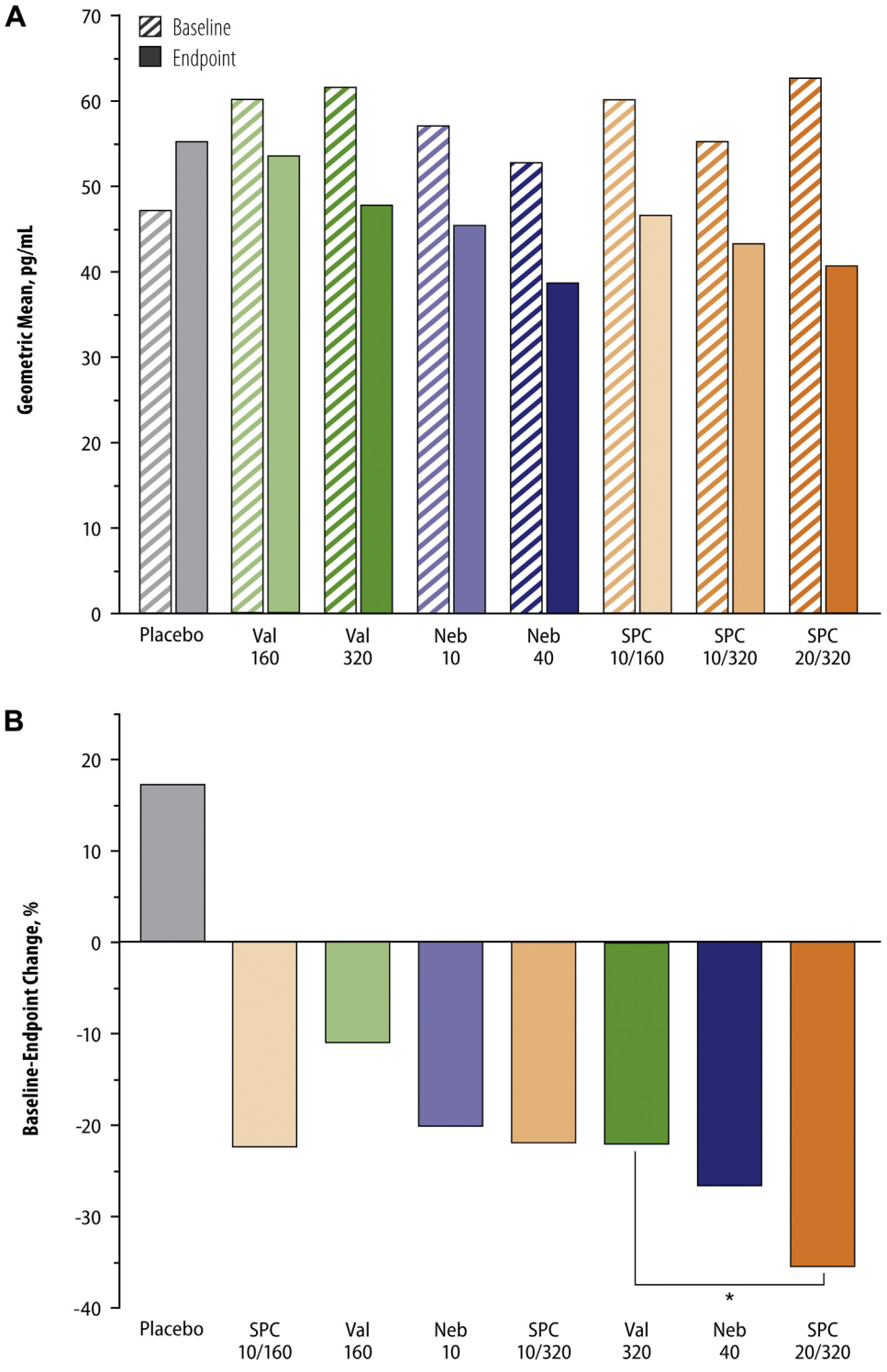

Figure 3. Aldosterone levels and changes from baseline (ITT, LOCF) (A) Baseline and Endpoint Concentrations; (B) Baseline-to-Endpoint Change. *$P < .05$. ITT, intent-to-treat population; LOCF, last observation carried forward; Neb, nebivolol; PRA, plasma renin activity; SPC, single-pill combination of nebivolol and valsartan; Val, valsartan.

There is potential importance in the suppression of the RAAS by the combination of nebivolol and valsartan. In fact, the decrease in PRA is regarded as a primary mechanism of BP lowering with β-blockers.[13] Results of our study cast light on results of another trial, in which patients with compromised renal function were treated

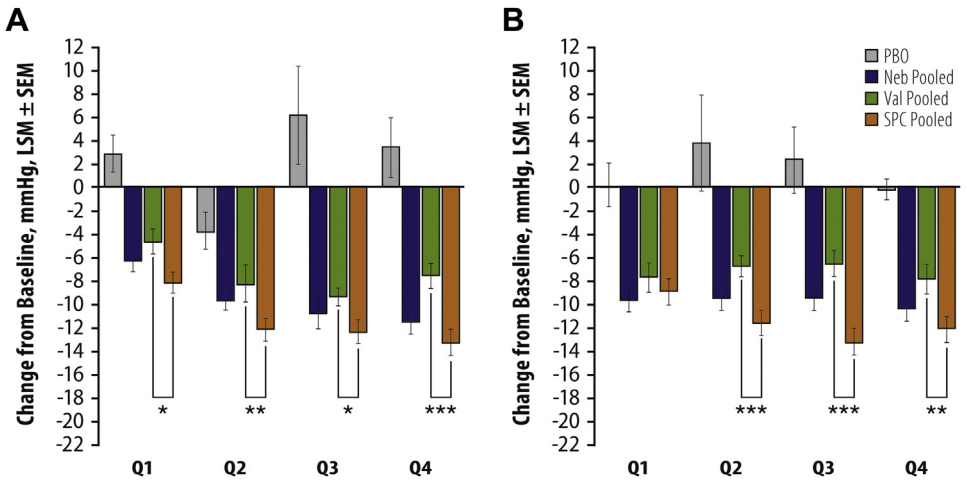

Figure 4. Baseline-to-endpoint changes in 24-hour DBP by baseline PRA and ALDO quartiles, pooled treatment groups (ITT, LOCF, ANCOVA) (A) 24-h DBP by PRA Quartile; (B) 24-h DBP by ALDO Quartile. *$P < .05$; **$P < .01$; ***$P < .001$. ABPM, ambulatory blood pressure monitoring; ALDO, aldosterone; ANCOVA, analysis of covariance; DBP, diastolic blood pressure; ITT, intent-to-treat population; LOCF, last observation carried forward; LS, least squares; SBP, systolic blood pressure; SEM, standard error of the mean; Q, quartile.

Table 1
Demographic and clinical characteristics at baseline, biomarkers-ABPM substudy (ITT population)

Characteristic	Placebo	SPC, Final Dosage (mg/day)			Nebivolol, Final Dosage (mg/day)		Valsartan, Final Dosage (mg/day)	
	(n = 52)	10/160 (n = 107)	10/320 (n = 108)	20/320 (n = 110)	10 (n = 105)	40 (n = 107)	160 (n = 103)	320 (n = 105)
Age (years)*	49.4 ± 11.1	51.5 ± 10.0	51.0 ± 8.7	50.7 ± 10.2	51.5 ± 9.1	51.4 ± 10.8	52.2 ± 9.6	50.6 ± 9.1
Men†	32 (61.5)	53 (49.5)	65 (60.2)	67 (60.9)	62 (59.0)	63 (58.9)	56 (54.4)	63 (60.0)
Race†								
White	36 (69.2)	84 (78.5)	90 (83.3)	92 (83.6)	85 (81.0)	92 (86.0)	88 (85.4)	84 (80.0)
Black	13 (25.0)	19 (17.8)	14 (13.0)	14 (12.7)	18 (17.1)	14 (13.1)	12 (11.7)	15 (14.3)
Other	3 (5.8)	4 (3.7)	4 (3.7)	4 (3.6)	2 (1.9)	1 (0.9)	3 (2.9)	6 (5.7)
Ethnicity†								
Hispanic	18 (34.6)	46 (43.0)	40 (37.0)	42 (38.2)	31 (29.5)	36 (33.6)	33 (32.0)	35 (33.3)
Non-Hispanic	34 (65.4)	61 (57.0)	68 (63.0)	68 (61.8)	74 (70.5)	71 (66.4)	70 (68.0)	70 (66.7)
Weight (kg)*	95.6 ± 22.7	91.9 ± 19.0	93.4 ± 17.6	92.8 ± 21.0	94.4 ± 20.2	92.5 ± 22.2	92.4 ± 19.4	94.8 ± 20.4
BMI (kg/m^2)*	32.5 ± 5.8	32.4 ± 6.0	32.4 ± 5.8	32.0 ± 6.4	32.4 ± 5.5	32.0 ± 6.2	32.2 ± 5.6	32.9 ± 6.2
Type 2 diabetes†	7 (13.5)	14 (13.1)	21 (19.4)	19 (17.3)	14 (13.3)	17 (15.9)	17 (16.5)	15 (14.3)
ABPM Values								
24-h SBP (mmHg)*,‡	137.3 ± 14.9	142.9 ± 14.3	142.8 ± 13.4	142.2 ± 13.5	138.9 ± 10.6	140.8 ± 13.9	142.6 ± 14.6	141.8 ± 13.1
24-h DBP (mmHg)*,‡	85.8 ± 9.6	88.1 ± 9.3	88.7 ± 9.1	87.3 ± 9.0	86.9 ± 8.6	86.6 ± 9.3	87.8 ± 9.6	88.6 ± 9.3

ABPM, ambulatory blood pressure monitoring; BMI, body mass index; DBP, diastolic blood pressure; ITT, intent-to-treat population; SD, standard deviation; SBP, systolic blood pressure; SPC, single-pill combination of nebivolol-valsartan.

ABPM values are based on the numbers of patients who had measurements at baseline and at least one postbaseline visit (placebo, n = 39; SPC 10/160, n = 78; SPC 10/320, n = 83; SPC 20/320, n = 86; nebivolol 10, n = 87; nebivolol 40, n = 77; valsartan 160, n = 78; valsartan 320, n = 82).

* Data presented as mean ± SD.

† Data presented as n (%).

‡ Intent-to-treat population.

Table 2
Correlation between baseline biomarker values and BP changes from baseline at week 8, pooled treatment groups (ITT, LOCF)

	ln(PRA)	ln(ALDO)
	Pearson's *R* *P*-Value	Pearson's *R* *P*-Value
24-hour DBP change		
Pbo	.113	.066
	.511	*.704*
Neb pooled	−.331	.003
	<.001	*.974*
Val pooled	−.101	−.073
	.216	*.374*
SPC pooled	−.209	−.181
	<.001	***.005***
24-hour SBP change		
Pbo	.034	.120
	.842	*.486*
Neb pooled	−.344	−.016
	<.001	*.844*
Val pooled	.009	−.063
	.908	*.439*
SPC pooled	−.189	−.217
	.003	***<.001***
Daytime DBP change		
Pbo	.157	.020
	.360	*.906*
Neb pooled	−.332	−.012
	<.001	*.878*
Val pooled	−.118	−.127
	.146	*.121*
SPC pooled	−.232	−.186
	<.001	***.004***
Daytime SBP change		
Pbo	.080	.051
	.641	*.768*
Neb pooled	−.330	−.013
	<.001	*.870*
Val pooled	−.002	−.109
	.984	*.181*
SPC pooled	−.208	−.225
	.001	***<.001***
Nighttime DBP change		
Pbo	−.055	.167
	.749	*.331*
Neb pooled	−.207	.029
	.008	*.720*
Val pooled	−.032	.056
	.693	*.496*
SPC pooled	−.103	−.122
	.108	*.061*
Nighttime SBP change		
Pbo	−.104	.256
	.546	*.132*
Neb pooled	−.279	−.020
	<.001	*.800*
Val pooled	.031	.049
	.709	*.551*
SPC pooled	−.107	−.151
	.094	***.020***

All *P*-values are presented as italic, and those <.05 are bold, as intended.

ABPM, ambulatory blood pressure monitoring; ALDO, aldosterone; BP, blood pressure; DBP, diastolic blood pressure; SPC, single-pill combination (nebivolol-valsartan fixed-dose combination); ITT, intent-to-treat population; ln, natural logarithm; LOCF, last observation carried forward; Neb, nebivolol; Pbo, placebo; SBP, systolic blood pressure; Val, valsartan.

with a combination of two RAAS blockers that increase renin release: aliskiren, a direct renin inhibitor, and valsartan.[14] The aliskiren-valsartan combination did not result in a favorable clinical response, perhaps due to an excessive increase in PRA,[15,16] an effect that contrasts strongly with the reduction in PRA observed with dual RAAS blockade in the present study. In fact, an increase in PRA may signal an unfavorable cardiovascular outcome, independent of any BP reduction.[17,18] It is entirely possible that the beta blocker-ARB combination we studied is a desirable approach to dual blockade of the renin-aldosterone axis.

The effect of the nebivolol-valsartan combination on aldosterone appears to be more complex. Nebivolol monotherapy has been shown to reduce plasma aldosterone levels in individuals with hypertension,[10] which is consistent with our data. Reductions in aldosterone also occur with valsartan treatment, but a phenomenon known as aldosterone ''breakthrough'' or ''escape''—a return toward pre-RAAS inhibitor treatment levels after an initial decrease[1,19]—has been observed in a subset of patients receiving ARB or angiotensin I-converting enzyme inhibitor treatment.[19] This aldosterone rebound with ARB treatment might be due to the inhibition of the angiotensin II-activated negative feedback loop, resulting in increased angiotensinogen synthesis.[1] In our study population, however, aldosterone levels decreased with valsartan monotherapy, at least during the 8-week period of observation. Indeed, all active treatments in our study were associated with a decrease in aldosterone levels, with the greatest effects during combination therapy, suggesting a potential combined effect. In addition, due to their opposing effects on renin release, nebivolol and valsartan given in combination might mitigate aldosterone breakthrough, if indeed this phenomenon occurs. However, such a possibility would have to be tested in appropriately designed clinical trials.

Perhaps the most interesting finding in this study—based on a post hoc analysis—was that the amplitude of the

BP-lowering effects of the combined nebivolol and valsartan treatment correlated with baseline values for both PRA and aldosterone. The finding with PRA, as discussed earlier, has been reported with other therapies, though usually with just one drug. The finding with aldosterone, however, appears to be unique, and underscores that this type of dual RAAS blockade might exert an important part of its effects through inhibition of aldosterone in patients whose hypertension may be at least partly dependent on the direct BP-raising effects of aldosterone. If so, this mechanism of action may be of importance in the treatment of patients with so-called treatment-resistant hypertension, in whom excess aldosterone may be an important factor in sustaining high BP levels.[20] It would be of considerable interest to prospectively measure the antihypertensive efficacy of the nebivolol-valsartan combination in such patients.

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Appendix. NAC-MD-01 Substudy Investigators

John Sorin, MD; Diverse Clinical Research Center Of Chicago
Matthew Davis, MD; Rochester Clinical Reseaerch, Inc.
Joesph Izzo, Jr., MD; Erie County Medical Center
Nabile Andrawis, MD; Manassas Clinical Research Center
Alyn Anderson, MD; Clinical Innovations, Inc.
Rogelio Bardinas-Rodriguez, MD; Sanitas Research
Douglas Young, MD; Northern California Research
Andrew Schreiber, MD; SDS Clinical Trials, Inc.
Cristian Breton, MD; International Research Associates, LLC
Duane Harris, MD; Intermountain Clinical Research
Phillip LaStella, MD; TKL Research, Inc.
Ramon Castello, MD; Jacksonville Center for Clinical Research
Susan Hole, DO; Riverside Clinical Research
Joesph Lillo, DO; Elite Clinical Studies
Luis Carlos Quintero, MD; International Research Associates, LLC
Carlos Montenegro, MD; MCS Clinical Trials
Jeffrey Rosen, MD; Clinical Research of South Florida
Farid Marquez, MD; Palm Springs Research Institute
Fredric Adler, MD; Olive View - UCLA Medical Center
Sady Alpizar, MD; Clinical Research Trials of Florida
James Andersen, MD; Meridien Research
Corey Anderson, MD; Dedicated Clinical Research
Graciela Calatayud, MD; Del Rosario Medical Clinic
Kevin Cannon, MD; PMG Research of Wilmington, LLC
Deanna Cheung, MD; Long Beach Center for Clinical Research
Rafel Chiong, MD; Kendall South Medical Center
Lisa Cohen, DO; Suncoast Clinical Research
Harry Collins, MD; Anderson & Collins Clinical Research, Inc.
Michael Dao, MD; SC Clinical Research
Cara H. Dawson, MD; Horizons Clinical Research Center
Donna DeSantis, MD; Clinical Research Advantage/East Valley Family Physicians, PLC
Shelly Dunmyer, MD; Pharmacotherapy Research Associates, Inc.
Sherif El-Harazi, MD; Global Reseach Management
Cecil M. Farrington, MD; PMG Research of Salisbury
David Ferrera, MD; Superior Research
Gregory S. Funk, DO; Fundamental Research
Gregory Gottschlich, MD; New Horizons Clinical Research
Terence T. Hart, MD; Terence Hart, MD
Marvin Kalafer, MD; The Clinical Trial Center, LLC
Dean Kereiakes, MD; The Linder Center
Gigi Lefebvre, MD; Meridien Research- St. Petersburg
Aristolis Laliotis, MD; Integrated Research Center
Peter Mattar, MD; St. Mark Clinical Trials, LLC
Michael McCartney, MD; ActivMed Practices & Research, Inc.
Diane McConnehey, DO; Northwest Clinical Trials Inc.
Curtis Mello, MD; Infinity Medical Research
Joel Neutel, MD; Orange County Research Center
Deborah A. Burke, MD; Comprehensive Clinical Development, Inc.
James Pritchard, MD; Sentral Clinical Research Services
George Raad, MD; PMG Research of Charlotte
Bruce Rankin, DO; Avail Clinical Research
John "Chip" H. Reed, MD; Endocrine Research Solutions Inc.
Erich Schramm, MD; St. Johns Center for Clinical Research
Howard Schwartz, MD; Miami Research Associates
Nathan Segall, MD; Clinical Research Atlanta
James Shoemaker, DO; Ormond Medical Arts Pharmaceutical Research Center
Vakas Sial, MD; TriWest Research Associates
Teresa Sligh, MD; Providence Clinical Research
William Smith, MD; Volunteer Research Group
Richard Stewart, MD; Perimeter Institute for Clinical Research, Inc.
Dan Streja, MD; Infosphere Clinical Research, Inc.
Danny Sugimoto, MD; Cedar Crosse Research Center
Alexander White, MD; Progressive Medical Concepts
Hayes Williams, MD, PhD; Achieve Clinical Research, LLC
William Abraham, MD; Radiant Research, Inc.
Azazuddin Ahmed, MD; Apex Medical Research, Inc. (AMR)
Richard Beasley, MD; Health Concepts
Daniel Gruener, MD; CRI Worldwide, LLC
Connie Hsu, MD; Dedicated Clinical Research
Ryan Klein, MD; Southern California Clinical Trials
Allen Soo, MD; Agave Clinical Research, LLC
Charles P. Andrews, MD; Diagnostics Reasearch Group
Clinton Corder, MD, PhD; COR Clinical Research, LLC
Donald Hurley, DO; Medical Research South, LLC
Elizabeth Bretton, MD; Albuquerque Clinical Trials, Inc.
Richard Martinez, MD; South Texas Medical Research Inc.
David Morin, MD; Holston Medical Group, PC
Miguel Trevino, MD; Innovative Research of West Florida, Inc.
Samir Arora, MD; Columbus Clinical Research, Inc
Curtis Scott Horn, MD; Quality Research, Inc.
Charles Lovell, MD; Sentara Medical Group
Thomas Nussdorfer, MD; Medical Research Associates, Inc.
Robert Weiss, MD; Maine Research Associates
Harold Bays, MD; L-MARC Research Center
Jackson Rhudy, MD; Optimum Clinical Research, Inc.

Edwardo Almaguer, MD; Eastern Research, Inc.

Joseph H. Woolley, MD; Chrysalis Clinical Research

Vicki Miller, MD; Texas Center for Drug Development, Inc.

Jaynier Moya-Hechevarria, MD; In Vivo Clinical Research, Inc.

Henry Punzi, MD; Punzi Medical Center

Addison Taylor, MD, PhD; Baylor College of Medicine

Jonathan Wilson, DO; PMG Research of Winston-Salem

Arnold Alper, MD; New Orleans Center for Clinical Research

Patricia Buchanan, MD; Williamette Valley Clinical Studies

Richard Dobrusin, DO; Clinical Research Advantage/ Mesa Family Medical Center, PC

Alan Forker, MD; St. Luke's Lipid & Diabetes Research Center

Razmig Krumian, DO; Westlake Medical Research

Samuel F. Oberstein, MD; Clinical Pharmacology of Miami, Inc.

Andrew Lewin, MD; National Research Institute

Mary Bella Natividad, MD; Millenium Clinical Trials, LLC

Armando Segui, MD; Elite Research Institute, Inc.

Wayne Harper, MD; Wake Research Associates, LLC

Andrea Lawless, MD; Biofortis Clinical Research

Lawrence S. Levinson, MD; Tipton Medical & Diagnostic Center

Shaukat Shah, MD; St. Joseph Medical Associates

Loray Blair-Britt, MD; Century Clinical Research, Inc.

Patrick Carmichael, MD; Florida Research Network, LLC

Thomas D. Giles, MD; Gulf Regional Research & Educational, Svc., LLC

Nathaniel Winer, MD; SUNY Downstate Medical Center

David Grant, MD; Sun Research Institute, LLC

Kyle Rickner, MD; INTEGRIS Family Care Yukon

Absalom Tilley, MD; Clinical Trials of America - Pinnacle Physicians Group

Linda Harper, MD; Clinical Neuroscience Solutions, Inc.

Stephen Maddock, MD, PhD; Analab Clinical Research, Inc.

Joseph A. Boscia, III, MD; CU Pharmaceutical Research

Yekaterina Khronusova, MD; Advanced Biomedical Research of America

Larry D. Reed, PhD, MD; HealthCare Research

Chandar Abboy, MD; Greenville Pharmaceutical Research